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Opti-Clot Study Grp

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ORIGINAL ARTICLE

Population pharmacokinetics of factor IX in hemophilia B patients undergoing surgery

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Essentials

- Factor IX (FIX) dosing using body weight frequently results in under and overdosing during surgery.
- We aimed to establish a population pharmacokinetic (PK) model describing the perioperative FIX levels.
- Population PK parameter values for clearance and V_1 were $284 \text{ mL h}^{-1} 70 \text{ kg}^{-1}$ and $5450 \text{ mL } 70 \text{ kg}^{-1}$.
- Perioperative PK parameters differ from those during non-surgical prophylactic treatment.

Summary. *Background:* Hemophilia B is a bleeding disorder characterized by a deficiency of coagulation factor IX (FIX). In the perioperative setting, patients receive FIX concentrates to ensure hemostasis. Although FIX is usually dosed according to bodyweight, under- and overdosing occurs frequently during surgery. *Aim:* The

objective was to quantify and explain the interpatient variability of perioperatively administered plasma-derived (pd) and recombinant (r) FIX concentrates. *Methods:* Data were collected from 118 patients (median age, 40 years [range, 0.2–90]; weight, 79 kg [range, 5.3–132]) with moderate (28%) or severe hemophilia B (72%), undergoing 255 surgical procedures. Population pharmacokinetic (PK) parameters were estimated using nonlinear mixed-effect modeling in NONMEM. *Results:* Measured perioperative FIX level vs. time profiles were adequately described using a three-compartment PK model. For a typical 34-year-old patient receiving rFIX, clearance (CL), intercompartmental clearance (Q_2 , Q_3), distribution volume of the central compartment (V_1) and peripheral compartments (V_2 , V_3) plus interpatient variability (%CV) were: CL, $284 \text{ mL h}^{-1} 70 \text{ kg}^{-1}$ (18%); V_1 , $5450 \text{ mL } 70 \text{ kg}^{-1}$ (19%); Q_2 , $110 \text{ mL h}^{-1} 70 \text{ kg}^{-1}$; V_2 , $4800 \text{ mL } 70 \text{ kg}^{-1}$; Q_3 , $1610 \text{ mL h}^{-1} 70 \text{ kg}^{-1}$; V_3 , $2040 \text{ mL } 70 \text{ kg}^{-1}$. From 0.2 years, CL and V_1 decreased 0.89% and 1.15% per year, respectively, until the age of 34 years. Patients receiving pdFIX exhibited a lower CL (11%) and V_1 (17%) than patients receiving rFIX. Interpatient variability was successfully quantified and explained. *Conclusions:* The estimated perioperative PK parameters of both pdFIX and rFIX are different from those reported for prophylactic treatment. The developed model may be used to apply PK-guided dosing of FIX concentrates during surgery.

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Introduction

Hemophilia B is a bleeding disorder characterized by a deficiency of coagulation factor IX (FIX). Severe and moderate patients have endogenous FIX levels less than 0.01 IU mL^{-1} and between 0.01 and 0.05 IU mL^{-1} , respectively [1,2]. In this category of patients, plasma-derived FIX (pdFIX) or recombinant FIX (rFIX) standard half-life concentrates are usually administered prophylactically to prevent spontaneous joint and muscle bleedings [3,4] and 'on-demand' when bleeding occurs in the surgical setting. In the prophylactic setting, FIX trough levels above 0.01 IU mL^{-1} are usually aimed for, as moderate patients have significantly fewer spontaneous bleeds [5]. In the perioperative setting, higher doses of FIX concentrates are administered to normalize FIX levels for 7–10 consecutive days post-surgery, with target trough levels from 1.00 to 0.30 IU mL^{-1} ensuring adequate hemostasis [6].

Currently, prophylactic, 'on-demand' and perioperative dosing of FIX concentrates is performed according to bodyweight with frequent monitoring to ensure sufficient FIX levels. Despite weight-based dosing, considerable under- and overdosing in the surgical setting has been reported by Hazendonk *et al.* [7]. It was shown that 60% of hemophilia B patients have FIX levels below the target level range during the first 24 h directly after surgery. This lack of adequate FIX plasma levels confers a considerable potential risk of bleeding and should be avoided. Therefore, more optimal dosing strategies are warranted.

In the prophylactic setting, FIX doses can be tailored to an individual's need by pharmacokinetic (PK)-guided dosing using Bayesian analysis [8]. In this approach, observed individual FIX levels are combined with PK information assessed in the population in order to obtain estimates for individual PK parameters [9]. These individual parameter estimates can be used to calculate doses necessary to achieve and maintain desired target levels by PK-guided dosing, potentially preventing over- and under-dosing. This approach can be applied iteratively, because with every new blood sample the calculated dose can be adapted to alterations in the individual PK parameter estimates [10]. This technique may also be applied in the perioperative setting.

A prerequisite for applying Bayesian analysis to perioperative dosing of FIX is the availability of a population model that describes the PK of FIX in hemophilia B patients undergoing surgery. The population PK of pdFIX and rFIX is well documented [5,11–13]. However, these models have all been constructed using data during non-surgical dosing of FIX concentrates. In the perioperative setting, the PK of FIX may, however, be altered. In order

to apply Bayesian dosing in the perioperative setting, a dedicated population model should be available.

This study was performed to describe the population PK of pdFIX and rFIX concentrates in hemophilia B patients during surgery and the days thereafter. It was investigated whether specific patient and surgical characteristics explain interpatient variability (IIV) in FIX exposure and whether the perioperative PK of FIX is similar to the prophylactic situation.

Methods

Patients and clinical data

An international multi-center observational cohort study was performed in which data were collected from 118 severe and moderate hemophilia B patients from five Hemophilia Treatment Centers in the Netherlands and five in the United Kingdom. Patients of all ages, who had undergone a minor or major elective surgical procedure between 1 January 2000 and 1 December 2015, were included [14]. Details of the study data have been reported previously [7].

In summary, severe and moderate hemophilia B patients received replacement therapy during surgery with FIX concentrates according to national and/or hospital guidelines, while aiming for target FIX levels as prescribed. To ensure hemostasis during the surgical procedure, a pdFIX product (AlphaNine[®] SD [Grifols Biologicals Inc., Los Angeles, CA, USA], Replenine[®] [Bio Products Laboratory, Elstree, UK], Haemonine[®] [Biotest Pharma GmbH, Dreierich, Germany], Mononine[®] [CSL Behring GmbH, Marbourg, Germany] and Nonafact[®] [Sanquin, Amsterdam, the Netherlands]) or rFIX product (BeneFix[®] [Pfizer Wyeth Pharmaceuticals Inc., Kent, UK] and IXinity[®] [Aptevo BioTherapeutics LLC, Berwyn, USA]) was administered with a bolus infusion of approximately 100 IU kg^{-1} , followed by either multiple intermittent bolus infusions or continuous infusions. FIX levels were obtained in the participating centers using a one-stage assay, according to local protocol.

Pharmacokinetic modeling

In population PK modeling, the PK is assessed in a cohort of patients rather than in an individual patient [15]. In population PK modeling, not only the average or median value of a PK parameter is of interest but also its inter- and intra-patient variability. Population PK parameters can be obtained by the standard two-stage method, in which individual PK parameters are calculated and, subsequently, summarized. A drawback of this method is that for each individual 10 or more serial samples should be available (rich sampling). In the clinical situation, this is often impossible or inconvenient to perform, especially

in populations such as children or the elderly. An alternative is the population approach [16], which allows the estimation of population PK parameters by analyzing data from all the patients simultaneously. The simultaneous analysis allows the use of sparsely and heterogeneously sampled data, which are frequently encountered in the clinical situation. In this study, sparsely and heterogeneously sampled data were used to construct the population PK model.

Using the population-based approach, a structural PK model is established first. This model consists of a number of PK compartments, with PK parameters described in terms of clearance and volume of distribution. The structural model provides values for the typical (average) parameter and, importantly, several levels of variability. Differences in PK parameters between patients are quantified in terms of interpatient variability (IIV). Variability of a PK parameter within a patient may be quantified by estimation of inter-occasion variability (IOV). Furthermore, a population PK model contains residual unexplained variability, which is the variability of the differences between the predicted and the measured plasma levels. By combining observed individual FIX levels and population PK parameters, empirical Bayesian estimates of the individual PK parameters can be obtained. These empirical Bayesian estimates can be used in the covariate analysis (below).

In this study, nonlinear mixed-effects modeling was used to estimate the population PK parameters [17]. A detailed description of the methods used to construct the population PK model can be found in Data S1. For each patient, the (historically lowest) endogenous baseline level was subtracted from each observed FIX level. Furthermore, in some subjects, a preoperative FIX level was present that was higher than the measured endogenous baseline level and for which no prior dose information was known. In the modeling procedure, the preoperative level was accounted for by an arbitrary virtual dose of 8250 IU administered 5 days prior to the pre-dose FIX measurement. To account for inter- and intra-individual variability in the observed preoperative FIX levels, the typical bioavailability of this dose was estimated in combination with its IIV and IOV. For estimation of the IOV, an occasion was defined as a single surgical procedure.

After the structural model was established, it was evaluated whether patient and surgical characteristics (covariates) explained the variability (IIV, IOV and residual unexplained variability) in a covariate model. Because FIX levels were available for both children and adults, estimated PK parameters were normalized for a bodyweight of 70 kg using allometric scaling with the $\frac{3}{4}$ rule [18]. Bodyweight was, however, missing in 38 surgical procedures (14.9%) involving 18 patients (15.3%). Therefore, a piecewise linear model was developed to impute the missing values for bodyweight using age as a predictor. Covariate relationships were evaluated using

graphical evaluation of plots of the empirical Bayesian estimates vs. the covariate value. Subsequently, covariates were implemented in the population model and their ability to explain the IIV, IOV or residual unexplained variability was tested by univariate analysis. The following covariates were evaluated: severity of hemophilia (severe vs. moderate), age, the use of tranexamic acid or heparin during surgery, the type of FIX concentrate (plasma derived or recombinant), the brand of product, treatment center, country of treatment, presence of hepatitis C, the use of prophylaxis before the surgical procedure, a history of neutralizing inhibitors, having a minor or major surgical procedure, blood group and the presence of an infection or a decrease in hemoglobin concentration during the surgical procedure. The final model, containing multiple covariates, was constructed by multivariate analysis using forward inclusion and backward deletion.

Model evaluation

The objective function value (OFV), which represents the ability of the model to describe the observed FIX levels, was used to discriminate between different models. When comparing nested models, the difference of the corresponding OFVs (dOFV) is known to be described by a chi-squared distribution, in which the difference in the number of parameters between the evaluated models determines the degrees of freedom. Therefore, a dOFV bigger than 3.84, 5.99 or 7.81 indicates a significant difference of $P < 0.05$ with 1, 2 or 3 degrees of freedom, respectively. In the covariate analysis, covariates were selected in the forward inclusion and backward elimination procedure if dOFVs bigger than 3.84 ($P < 0.05$, d.f. = 1) and 6.63 ($P < 0.01$, d.f. = 1), respectively, were obtained.

To evaluate whether the measured FIX levels were adequately described by the developed population PK model, several criteria were used. The adequacy of the model was evaluated by inspection of precision of the estimated model parameters, creation of goodness-of-fit plots, evaluation of shrinkage of the IIV, IOV and residual unexplained variability, the condition number of the model and the creation of visual predictive checks [19,20]. In the latter procedure, FIX levels were generated by Monte Carlo simulation ($n = 1000$) using the established population PK model and are, subsequently, compared to the actual measured FIX levels [21]. For the goodness-of-fit plots, the measured FIX levels were compared with the population predicted FIX levels using the typical values for the PK parameters and the individual FIX levels predicted on basis of the empirical Bayesian estimate. Moreover, several plots were evaluated depicting conditional weighted residuals (CWRES). CWRES are the weighted difference between the model-predicted and measured FIX levels [22].

The stability and robustness of the final model were tested by a bootstrap analysis [23]. In this analysis, 1000

new datasets were created by randomly sampling from the data from all patients in the original dataset. Subsequently, the final model was re-estimated using the bootstrapped datasets. The median and 95% confidence interval of the obtained bootstrap parameters were compared with the estimated PK parameters of the final model.

Comparison with non-surgical FIX models

The final population model describing the PK of FIX in hemophilia B patients during surgery was compared with published population PK models derived from data of patients on prophylaxis [11–13,24]. To evaluate whether the published prophylactic models were able to describe the perioperative FIX levels from this study, predictions of the perioperative FIX levels were calculated using the prophylactic population PK parameters. For each model, the difference between the population predictions and the measured FIX levels was summarized using the relative mean prediction error (rMPE). The latter was calculated using the following equation:

$$\text{rMPE} = \frac{1}{n} \sum_{i=1}^n \left(\frac{C_{\text{pred}} - C_{\text{FIX:C}}}{C_{\text{FIX:C}}} \right) \times 100\% \quad (1)$$

in which C_{pred} are the population predictions and $C_{\text{FIX:C}}$ the measured FIX level for a total of n measurements. Furthermore, the terminal elimination half-life was calculated using the values from all population PK parameters.

Results

Patients

In total, 118 severe and moderate hemophilia B patients were included, undergoing 262 surgical procedures. Four occasions were excluded, as FIX levels were not measured. Because of the withdrawal of approval for IXinity® by the European Medicine Agency during data collection in June 2013, another three surgical procedures were also excluded from analysis [25]. As a result, data from 255 surgical procedures were used for PK analysis. Table 1 shows the general patient characteristics.

Table 1 General characteristics of the study population

	Total cohort		Adults		Children*	
	<i>N</i> (%) or median [range]					
Patient characteristics						
No. of patients	118		82		36	
Age (years)	40	[0.2–90]	46	[18–90]	6	[0.2–18]
Bodyweight (kg)	79	[5.3–132]	85	[47–132]	19	[5.3–117]
Severe hemophilia B (< 0.01 IU mL ^{−1})	85	(72)	57	(70)	28	(78)
On prophylaxis	36	(31)	28	(34)	8	(22)
Blood group O†	33	(28)	24	(29)	9	(25)
Neutralizing antibodies (historically)	6	(5)	5	(6)	1	(3)
Chronic hepatitis C	47	(40)	46	(56)	1	(3)
Patient treated in the UK	93	(79)	63	(77)	30	(83)
Surgical characteristics						
No. of surgical procedures	255		201		54	
Total no. of patients undergoing:						
1	118		82		36	
2	63		49		14	
3	32		28		4	
> 3	42		42		0	
Minor surgical procedures	135	(53)	96	(48)	39	(72)
Major surgical procedures	120	(47)	105	(52)	15	(28)
Replacement therapy with FIX concentrate						
Mode of infusion						
Continuous	56	(22)	54	(27)	2	(4)
Bolus	199	(78)	147	(73)	52	(96)
Product type						
Recombinant	201	(79)	150	(75)	51	(91)
Plasma derived	54	(21)	51	(25)	3	(6)
Pharmacokinetic data						
Total number of observations	1555		1324	(85)	231	(15)
No. of observations per occasion	4	[1–23]	10	[1–23]	5	[1–16]
No. of doses per occasion	7	[1–52]	12	[1–52]	12	[1–39]

*Children were defined as having an age less than 18 years. †Blood group available in 80 patients. Adapted from Hazendonk *et al.* [7] with permission.

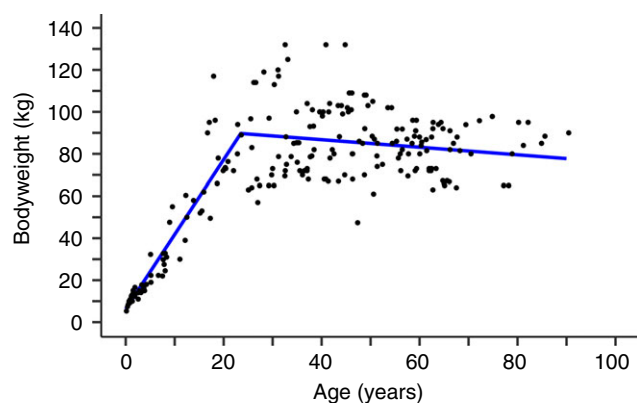


Fig. 1. Imputed bodyweight vs. age. Black dots are individual bodyweights for 99 patients. The blue line depicts the predicted bodyweight with age as a predictor, as described by the following piecewise linear model: $WT_{est} \text{ (kg)} = 6.3 + 3.6 \times AGE - 3.73 \times (AGE - 23.5)^{DAGE}$. In this equation, WT_{est} is the estimated bodyweight and $DAGE$ is 1 in the case that the age of the patient is 23.5 years or older; in every other case it is zero. [Color figure can be viewed at wileyonlinelibrary.com]

Bodyweight was not recorded in 14.9% of all surgical procedures. Therefore, a piecewise linear model was developed, from which the missing values for bodyweight could be imputed using age (Data S1). Table S1 shows the parameter estimates for the piecewise linear model. The relationship between age and bodyweight is shown in Fig. 1; the blue line depicts the predictions from the model for all ages, which was used to impute values for the missing bodyweights.

Pharmacokinetic modeling

For constructing the structural model, a three-compartment model more adequately fitted the FIX levels than a two-compartment model ($dOFV = 58.1$, $P < 0.001$) (Figure S1). Table 2 summarizes the parameter estimates of the structural model. For all estimated PK parameters, the imprecision of the estimated value was lower than 20%. A proportional residual error model was most appropriate to fit the data, as compared with an additive or combined residual error model. In the structural model, IIV could be estimated for both CL and $V1$, as well as a correlation for the IIV between the two parameters. Moreover, shrinkage values for the IIV of CL and $V1$ were lower than 20%, indicating that there was sufficient information available for each patient to estimate the individual parameters reliably [26]. Although IIV should also be present for the other PK parameters (e.g. $Q2$, $Q3$, $V2$, $V3$), the available data did not support the estimation of these values. Pre-administration FIX levels (greater than endogenous baseline values) were present in 138 of the 255 evaluated surgical procedures and ranged from 0.01 to 0.67 IU mL^{-1} . Administration of a virtual dose of 8250 IU, 120 h before the start of the surgery,

adequately approximated the pre-administration FIX levels and significantly improved the fit of the model; $dOFV$ was -495.5 ($P < 0.001$). The typical value for the estimated bioavailability of the virtual dose was 99.8% and IIV and IOV values were 91% and 93%, respectively. These values indicate that virtual doses vary largely between patients and surgical procedures, with values ranging from 744.2 to 196 824.2 IU. Estimation of IOV for differences in CL and $V1$ between surgical procedures was not successful. By implementing IOV for CL, the fit of the model improved ($dOFV$, -141.9 ; $P < 0.01$). However, parameter estimates became unstable and IOV was therefore not included.

Covariate analysis

To prevent the covariates influencing the estimation of the virtual dose, the IIV and IOV from the virtual dose were fixed to the values obtained for the structural model. Table 3 shows the $dOFV$ for the selected covariate relations from the forward inclusion and backward elimination procedure. Age of the patient was included for CL and $V1$ as a piecewise linear model, which is a linear model with two slopes. The best fit was obtained when the first slope was estimated and the second was set to zero from an age of 34 years, which was the median, and higher. As a result, the bodyweight-normalized CL and $V1$ decreased 0.89% and 1.15% per year, respectively, until the age of 34 years. Moreover, IIV was reduced from 20.8% to 18.5% (10.1%) and from 24.6% to 18.7% (14.6%) for CL and $V1$ as a result of the introduction of age. In Fig. 2(A,B), age vs. individual values for CL and $V1$, as obtained by Bayesian analysis using the final model, are shown. In these figures, the combined effect of bodyweight and age is observed, as CL and $V1$ increase with bodyweight and decrease with age up to 34 years.

Patients receiving pdFIX concentrates exhibited a lower CL and $V1$ as compared with patients receiving rFIX concentrates; respective values were 11% and 17% lower for pdFIX. Moreover, $V1$ was 10% lower in patients with moderate hemophilia in comparison to patients with severe hemophilia. The parameters of the final model are summarized in Table 2. All other covariate relations did not result in a significant $dOFV$.

Evaluation of the final model

The fit of the final model was evaluated by inspection of goodness-of-fit plots, as shown in Fig. 3(A–D). Figure 3(A) shows the prediction of FIX levels, based on the population PK parameter values, adjusted for the covariate values. Both under- and overprediction are present because IIV is not taken into account for calculating the population predictions. Nevertheless, the population predictions are distributed randomly around the $y = x$ axis, demonstrating the appropriateness of the model.

Table 2 Estimated population pharmacokinetic parameters for the structural model, final model and bootstrap analysis

	Structural model			Final model			Bootstrap analysis	
	Estimate	RSE (%)	Shr. (%)	Estimate	95% CI	Shr. (%)	Estimate	95% CI
Structural model								
Clearance (CL; mL h ⁻¹ 70 kg ⁻¹)	296	2.5		284	[266–302]		283	[265–300]
Volume of central compartment (V1; mL 70 kg ⁻¹)	5370	2.7		5450	[5005–5895]		5426	[4933–5886]
Distribution clearance to compartment 2 (Q2; mL h ⁻¹ 70 kg ⁻¹)	112	25		110	[86–134]		110	[92–140]
Volume of compartment 2 (V2; mL 70 kg ⁻¹)	4720	16.7		4800	[3793–5807]		4879	[4118–6367]
Distribution clearance to compartment 3 (Q3; mL h ⁻¹ 70 kg ⁻¹)	2210	19.9		1610	[– 32 to 3252]		1943	[667–5132]
Volume of compartment 3 (V3; mL 70 kg ⁻¹)	2160	17.7		2040	[1344–2736]		2079	[1376–2753]
Virtual dose	0.998	22.0		ND			ND	
Inter-individual variability (%CV)								
IIV on CL	20.8	9.1	12.3	18.5	[16.4–21.3]	10.2	18.5	[15.3–21.5]
IIV on V1	24.6	12.2	14.5	18.7	[16.5–21.4]	15.0	18.7	[14.0–23.5]
Correlation between CL and V1 (%)	91.3	11.2		89.4	[85.0–92.5]		89.1	[88.6–91.6]
IIV on virtual dose	94.5	18.0		ND			ND	
Inter-occasion variability (%CV)								
IOV on virtual dose	93.4	10.2		ND			ND	
Residual variability								
Proportional residual error (%CV)	23.0	4.2		21.9	[20.2–23.6]		21.7	[20.0–23.3]
Covariate relations								
CL (% change with age different from 34 years)				– 0.89	[– 1.4 to – 0.4]		–0.89	[– 1.4 to – 0.4]
V1 (% change with age different from 34 years)				– 1.15	[– 1.7 to – 0.6]		–1.14	[– 1.7 to – 0.6]
CL, plasma-derived product (%)				88.8	[83.8–93.8]		88.9	[84.3–94.6]
V1, plasma-derived product (%)				82.7	[74.7–90.7]		82.3	[72.5–90.8]
V1, if moderate hemophilia patient (%)				89.5	[82.8–96.2]		89.1	[82.3–96.3]
Model characteristics								
Objective function value	– 2827.12			– 2905.27			ND	
Condition number	68.65			119.65			ND	

CI, confidence interval as obtained using the 2.5th and 97.5th percentiles from the non-parametric distributions; CV, coefficient of variation; IIV, interpatient variability; IOV, inter-occasion variability; ND, not determined; RSE, relative standard error; Shr., shrinkage. The typical values are obtained for a severe hemophilia B patient weighing 70 kg receiving a recombinant factor IX product.

$$CL \text{ (mL h}^{-1}\text{)} = 284 \times \left(\frac{BW}{70}\right)^{0.75} \times (1 - 0.0089 \times (\text{Age} - 34))^{\text{AGE} < 34} \times 0.888^{\text{Plasma-derived product}}.$$

$$V1 \text{ (mL)} = 5450 \times \left(\frac{BW}{70}\right)^{1.0} \times (1 - 0.0115 \times (\text{Age} - 34))^{\text{AGE} < 34} \times 0.827^{\text{Plasma-derived product}} \times 0.895^{\text{moderate hemophilia}}.$$

Figure 3(B) is obtained after Bayesian analysis, in which individual PK parameter estimates are obtained by simultaneous analysis of the individual observations and the population model. The individual FIX levels are predicted using the derived empirical Bayesian estimates. Again, these predictions are distributed randomly around the $y = x$ axis. Figure 3(C,D) shows plots of the conditional weighted residuals (CWRES) vs. predicted FIX level and time, respectively. CWRES values are distributed randomly around the line $y = 0$ (see also Figure S3). Most of the values are within the -2 and $+2$ SD range, which confirms the goodness-of-fit of the final model.

To evaluate the stability of the final model, a bootstrap analysis was performed. In this analysis, 1000 model estimations were performed, from which 98.3% were successful. Table 2 shows that the medians for the parameter estimates from the bootstrap analysis were similar to those from the final model, except for Q3. This deviation for Q3 is caused by the high imprecision of its estimation, as shown by the 95% CI for Q3 from the bootstrap analysis (667.2–5131.9 mL h⁻¹70 kg⁻¹). For all other parameters of the final model, the CIs were small and corresponded to the relative standard errors from the parameter estimates of the final model.

Table 3 Model building-steps

		OFV	dOFV	No. of parameters
Structural model				
1	Structural model with doses calculated using the virtual dose	– 2827.1	ND	9
Covariate relationships – forward inclusion				
2	Structural model and age on CL*	– 2832.7	– 5.6	10
3	Model 2 and age on <i>V</i> ₁ *	– 2856.4	– 23.6	11
4	Model 3 and plasma-derived products on CL	– 2876.6	– 20.3	12
5	Model 4 and plasma-derived product on <i>V</i> ₁	– 2897.6	– 20.9	13
6	Model 5 and moderate patient, as compared with severe patient on <i>V</i> ₁	– 2905.3	– 7.7	14
Covariate relationships – backward deletion				
7	Model 6 without moderate patient, as compared with severe patient on <i>V</i> ₁	– 2897.6	7.7	13
8	Model 6 without age on CL	– 2881.4	23.9	13
9	Model 6 without age on <i>V</i> ₁	– 2883.2	22.1	13
10	Model 6 without plasma-derived products on CL	– 2871.8	33.5	13
11	Model 6 without plasma-derived products on <i>V</i> ₁	– 2880.3	25.0	13

OFV indicates objective function value, as calculated by minus two times the logarithm of the likelihood ($-2LL$) of the model describing the data; dOFV, difference of the corresponding OFVs; CL, clearance; ND, not determined; No., number; *V*₁, volume of distribution of the central compartment. *For these models, the coefficients for covariate age on both CL and *V*₁ were estimated using a piecewise linear model. However, the slope for the ages above 33.6 years was fixed to 0. Therefore, the number of parameters only increases by 1.

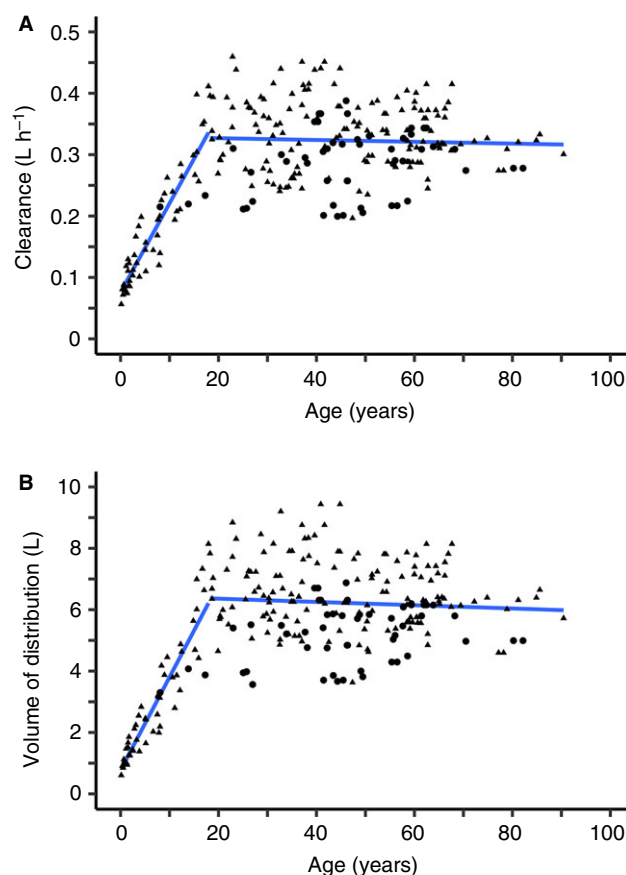


Fig. 2. Clearance and volume of distribution vs. age. The black dots depict the individual PK parameter estimates obtained by Bayesian analysis for the pdFIX (▲) and rFIX (●) data. (A) Clearance. (B) Volume of distribution of the central compartment. The blue line represents two (piecewise) linear fits of the data for patients having an age between 0.2–18 years and 18–90 years, respectively. CL and *V*₁ increase with bodyweight and decrease with age up to 34 years. In this figure, the combined effect of bodyweight and age is observed. Both parameters slightly decrease from the age of 18 years because of decreasing bodyweight with increasing age. [Color figure can be viewed at wileyonlinelibrary.com]

The evaluation of the final model comprised 1000 Monte Carlo simulations for each patient to construct a visual predictive check, as shown in Fig. 4 (and

Figure S4). The (grey) lines, depicting the 2.5th, 50th and 97.5th percentiles of the measured FIX levels, are predominantly within their corresponding 95% prediction

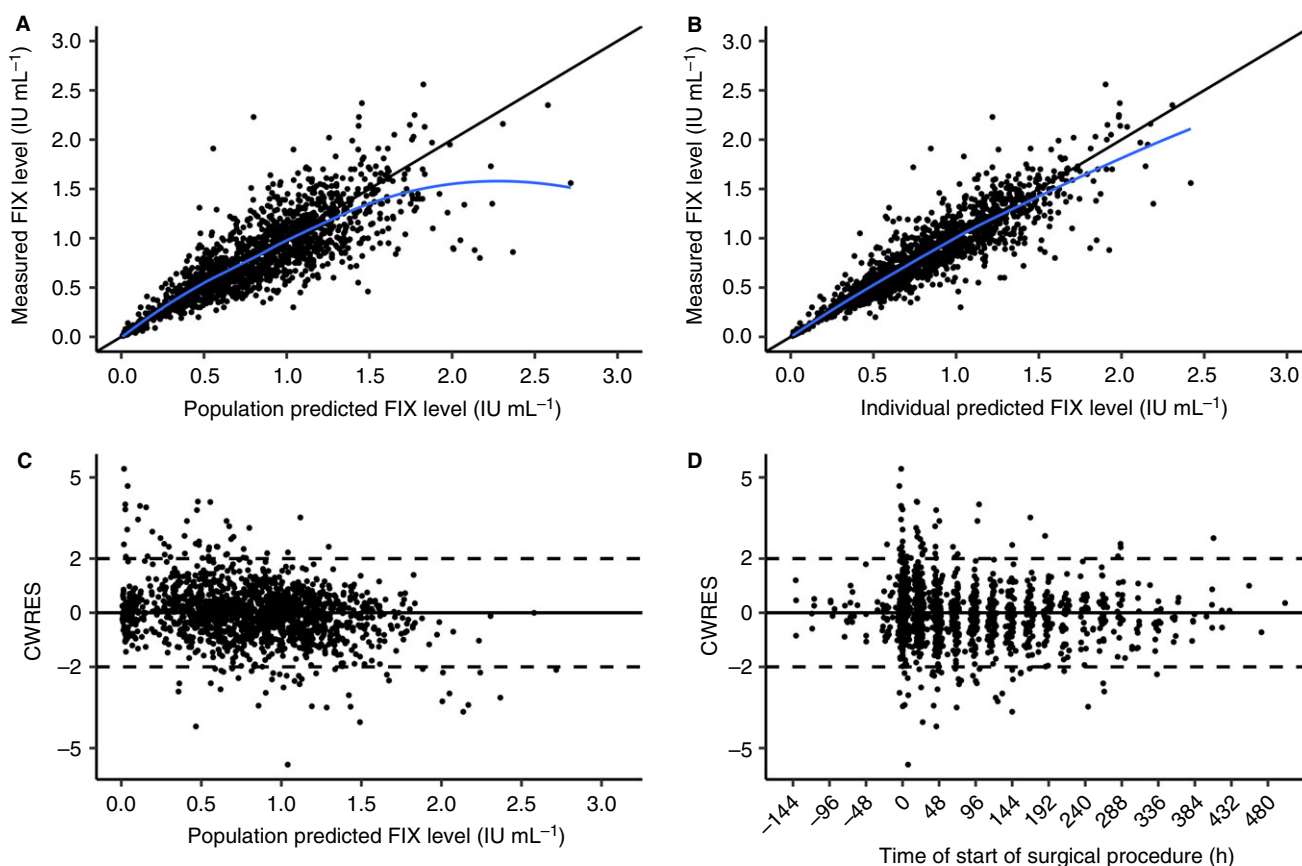


Fig. 3. Goodness-of-fit of the plot for the final model. (A) Population predicted vs. measured FIX levels. (B) Individual predicted vs. measured FIX levels. (C) Conditional weighted residuals (CWRES) vs. population predicted FIX levels. (D) CWRES vs. time, defined as the time of start of the surgical procedure. Negative times represent samples taken before the start of the surgical procedure. [Color figure can be viewed at wileyonlinelibrary.com]

intervals, as presented by blue and red areas. As a result, the simulated data were similar to the measured data, confirming the adequacy of the final model.

Comparison with non-surgical models

Table S2 summarizes population PK parameters of four models that have been published previously and were constructed using data obtained after non-surgical dosing. Higher values for the population PK parameter CL were found for the rFIX models as compared with the pdFIX models. Figure S2 shows the predicted FIX levels as obtained using the population PK parameter values analogous to Fig. 3(A) (without IIV). Figure S2(A,B) was constructed using solely the pdFIX data from this study; concentrations were predicted using the population parameters of pdFIX model 1 and 2. Likewise, Figure S2(C,D) was constructed using solely the rFIX data from this study in combination with population parameters from rFIX model 1 and 2. In each case, the non-surgical models underpredicted the observed levels, as shown by the blue lines being above the black line $y = x$. The rMPE values, calculated for pdFIX model 1

and 2 and rFIX model 1 and 2, were -7.2% , -15.7% , -40.7% and -40.3% , respectively. Furthermore, the half-lives calculated for pdFIX and rFIX using the population parameter values for the final model from the present study were 51 and 49 h, respectively, whereas the terminal elimination half-lives for pdFIX model 1 and 2 and rFIX model 1 and 2 were: 28, 23, 20 and 20.3 h, respectively.

Discussion

In this study, the PK of pdFIX and rFIX were characterized in children and adults with severe and moderate hemophilia B undergoing a surgical procedure. Considerable interpatient variability was identified for clearance and central volume of distribution, which was partially explained by the patient's age, type of FIX product and the severity of hemophilia. Importantly, the perioperative PK parameter of FIX was different from that in the non-surgical situation.

In population PK analysis, the variability within and between patients is quantified and, subsequently, explained using covariates such as age or bodyweight.

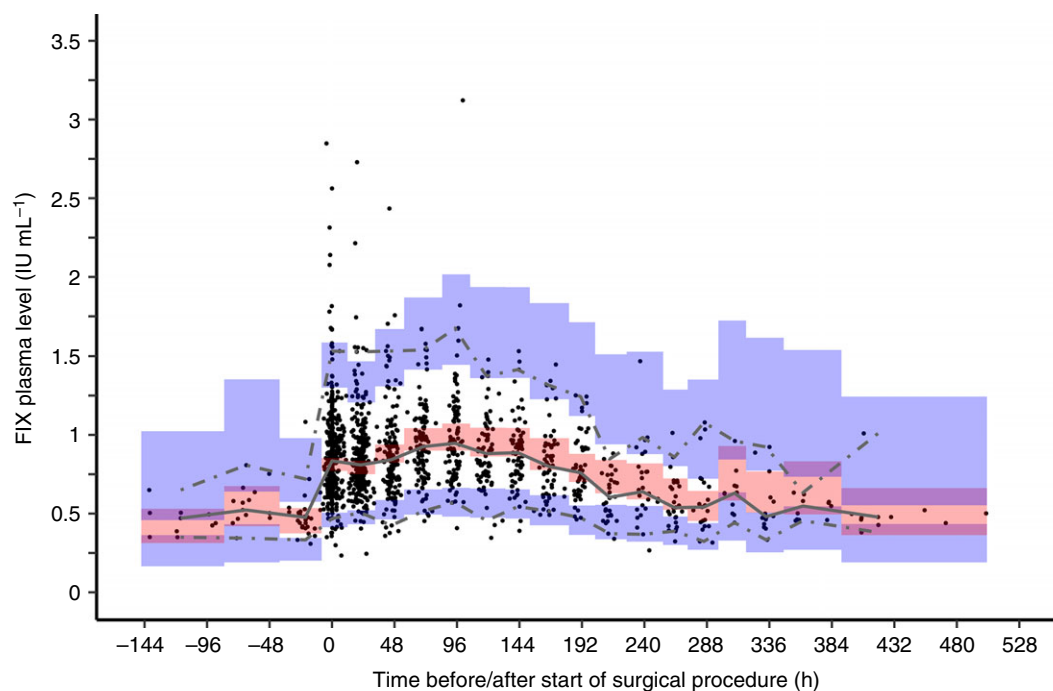


Fig. 4. Prediction-corrected visual predictive check of the final model. Time is defined as the time of start of the surgical procedure. Data with negative times represent samples taken before the start of the surgical procedure. Black dots represent the measured FIX levels for all patients. Solid grey line represents the median and the dashed grey lines represent the 2.5th and 97.5th quantiles of the measured FIX levels. Red and blue-shaded areas show the 95% confidence intervals for the predicted individual FIX levels, as obtained by 1000 Monte Carlo simulations using the final model. The binning of the areas for the prediction intervals was created using the auto-bin option in Perl-Speaks-NONMEM. [Color figure can be viewed at wileyonlinelibrary.com]

When these variabilities are assessed adequately, a population PK model may be used for PK-guided dosing using Bayesian analysis. In contrast to dosing based on bodyweight, PK-guided dosing allows for individualization of doses while taking the individual's PKs into account. To apply Bayesian analysis clinically, an appropriate population PK model is essential. Moreover, Bayesian analysis using a population PK model, which does not describe the PKs of FIX adequately, may result in biased individual PK parameters and, hence, biased estimated doses. For FVIII, a dedicated population PK model for hemophilia A patients undergoing a surgical procedure was constructed in a similar fashion [27]. Therefore, a dedicated population PK model was constructed to describe the perioperative FIX levels.

In this study, the observed presurgical FIX level was higher than the endogenous baseline value in 138 of 255 surgical procedures. These elevated presurgical FIX levels were taken into account by a virtual dose that was estimated using a typical value and both IIV and IOV. Thereby, each patient having a presurgical FIX level can have a different virtual dose for each surgical procedure. Inclusion of these presurgical FIX levels greatly improved the fit of the model. Therefore, exclusion of such presurgical FIX levels may lead to biased population PK parameter estimates.

In the present study, age partially explained the inter-individual variability from CL and V_1 . In the final model,

the best fit was obtained using a piecewise linear relation using two slopes with -0.89% and -1.15% for ages below 34 years for bodyweight-normalized CL and V_1 , respectively. Allometric scaling of CL using an exponential factor of 0.75 partly explains the increased clearance when a lower bodyweight is present. Nevertheless, additional variability was explained by taking age into account. Björkman *et al.* reported a similarly piecewise linear relationship between age and CL of rFIX when administered in a non-surgical situation [28,29]. It was shown that clearance and (steady-state) volume of distribution decreased when age increased from 2 to 20 years. Above an age of 20 years, there was virtually no change in clearance or volume of distribution. Suzuki *et al.* explored a similar piecewise relationship of age with CL for the population PKs of rFIX as well [13]. However, a relationship between age and CL could not be identified when bodyweight was included in the model as well. Furthermore, in the covariate analysis, severity of hemophilia B was associated with V_1 . For a moderate hemophilia B patient, V_1 was 10.5% lower as compared with a severely affected patient, which is in agreement with the findings from Ewenstein *et al.* [30].

In previous studies, differences have been reported between PK parameters from pdFIX and rFIX products in the non-surgical situation [5,30,31]. The *in vivo* recovery for rFIX products was found to be on average 53% that of pdFIX products [5]. As *in vivo* recovery is

inversely related to volume of distribution, V_1 is lower for pdFIX products. Moreover, the clearance of rFIX products was found to be approximately twice as high as compared with pdFIX products [32]. In the present study, CL and V_1 of pdFIX were 11.2% and 17.3% lower than their respective values for rFIX. These higher values for CL and V_1 from rFIX are in accordance with results from previous studies. However, the difference between the types of products is smaller in the surgical situation than in the non-surgical situation.

In Figure S2, each published non-surgical population PK model showed that the observed perioperative FIX levels were underpredicted. These differences were also demonstrated by simulations of the typical FIX level vs. time profiles for a patient receiving 100 IU kg⁻¹ of pdFIX or rFIX using the available population PK models (Figure S5). The calculated rMPEs and half-lives clearly demonstrate that the PK of FIX in the non-surgical setting is different from the surgical setting. The extent of underprediction was higher for rFIX model 1 and 2 (−40.7% and −40.3%) compared with pdFIX model 1 and 2 (−7.2% and −15.7%). This may be explained by the fact that CL in the non-surgical situation was almost twice as high as the value in the present study: 560 mL h⁻¹ 70 kg⁻¹ and 551 mL h⁻¹ 70 kg⁻¹ vs. 284 mL h⁻¹ 70 kg⁻¹, respectively. For the pdFIX models, there was less underprediction. Values for CL in the non-surgical situation were slightly higher than the values from the present study: 290 mL h⁻¹ 70 kg⁻¹ and 319.8 mL h⁻¹ 70 kg⁻¹ vs. 284 mL h⁻¹ 70 kg⁻¹. An explanation for this difference is unknown. Nevertheless, the currently published population PK models for prophylactic treatment with rFIX and pdFIX underpredict the perioperative FIX levels. Consequently, use of these models in the perioperative situation results in overdosing.

Conclusion

In the present study, a population PK model was established that adequately described the perioperative FIX levels obtained from hemophilia B patients undergoing a surgical procedure. As differences in the population PK parameters were found between the surgical and non-surgical setting, the dedicated population PK model constructed in this study may be applied for patient-tailored dosing in the perioperative period. However, application of a population PK model for clinical use should always be validated.

Addendum

T. Preijers and R. A. A. Mathôt performed the population PK analysis. T. Preijers, R. A. A. Mathôt and M. H. Cnossen wrote and revised the manuscript. M. H. Cnossen and R. A. A. Mathôt wrote the protocol and supervised the study in the Netherlands. M. H. Cnossen, H. C.

A. M. Hazendonk, P. W. Collins and R. Liesner wrote the protocol for the United Kingdom and organized the clinical study and data collection in the United Kingdom. H. C. A. M. Hazendonk performed data collection in all centers in both countries and analyzed the clinical data. The study protocol was implemented and patient inclusions were organized in the Netherlands by: H. C. A. M. Hazendonk, B. A. P. Laros-van Gorkom, F. J. M. van der Meer, K. Meijer, K. Fijnvandraat, F. W. G. Leebeek and M. H. Cnossen, and in the United Kingdom by H. C. A. M. Hazendonk, P. W. Collins, R. Liesner, P. Chowdary, D. Hart and D. Keeling. M. H. E. Driessens gave critical input at initiation of and during the clinical study. M. H. Cnossen, R. A. A. Mathôt, F. W. G. Leebeek and K. Fijnvandraat gave critical guidance during the study and for the population PK analysis. All authors contributed substantially to the writing and critical revision of the manuscript, and approved the final draft.

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Disclosure of Conflict of Interests

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from Pfizer, Baxalta/Shire, Bayer, CSL Behring, Novo Nordisk, Novartis, Nordic Pharma, and for advisory board activities from Bayer and Roche. R. Mathôt has received travel grants from Shire and Bayer. The remaining authors declare no competing financial interests.

Appendix

OPTI-CLOT Study Members

Steering Committee, the Netherlands: M. H. Cnossen (Principal Investigator and Chair) and F. W. G. Leebeek, Rotterdam; K. Fijnvandraat, R. A. A. Mathôt, Amsterdam. Principal investigators and local collaborators, the Netherlands: M. J. A. H. Kruip, S. Polinder, J. Lock, H. C. A. M. Hazendonk, I. van Moort, J. M. Heijdra, A. Nederlof, Rotterdam; R. A. A. Mathôt, K. Fijnvandraat, T. Preijers, N. de Jager, M. Coppens, M. Peters, Amsterdam; K. Meijer, R. Y. J. Tamminga, Groningen; P. Brons, B. A. P. Laros-van Gorkom, Nijmegen; F. J. M. van der Meer, H. C. J. Eikenboom, Leiden; R. E. G. Schutgens, K. Fischer, Utrecht; M. H. E. Driessens, Nijkerk. Website, trial bureau and databases: C. M. Zwaan, I. van Vliet, Rotterdam.

Principal investigators and local collaborators in the United Kingdom: P. W. Collins, Arthur Bloom, Cardiff; R. Liesner, P. Chowdary, D. Hart, London; D. Keeling, Oxford.

Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article:

Data S1. Methods.

Table S1. Model for bodyweight imputation using age

Table S2. Population PK parameter estimates from published models

Fig. S1. Depiction of a three-compartment PK model.

Fig. S2. Predictions of perioperative FIX levels using published population PK models.

Fig. S3. Conditional weighted residuals (CWRES) vs. time after dose.

Fig. S4. Prediction corrected visual predictive check.

Fig. S5. Simulated population predictions from the available FIX population PK models for a typical patient.

References

- Srivastava A, Brewer AK, Mauser-Bunschoten EP, Key NS, Kitchen S, Llinas A, Ludlam CA, Mahlangu JN, Mulder K, Poon MC, Street A; Treatment Guidelines Working Group on Behalf of The World Federation Of Hemophilia. Guidelines for the management of hemophilia. *Haemophilia* 2013; **19**: e1–47.
- Blanchette VS, Key NS, Ljung LR, Manco-Johnson MJ, van den Berg HM, Srivastava A; the Subcommittee on Factor VIII, Factor IX and Rare Coagulation Disorders. Definitions in hemophilia: communication from the SSC of the ISTH. *J Thromb Haemost* 2014; **12**: 1935–9.
- Leebeek FWG, Mauser-Bunschoten EP. Nieuwe richtlijn diagnostiek en behandeling van hemofilie en aanverwante hemostasestoornissen. *Ned Tijdschr Hematol* 2010; **7**: 107–14.
- United Kingdom Haemophilia Centre Doctors' Organisation. Guidelines on the selection and use of therapeutic products to treat haemophilia and other hereditary bleeding disorders. *Haemophilia* 2003; **9**: 1–23.
- Björkman S. Pharmacokinetics of plasma-derived and recombinant factor IX – implications for prophylaxis and on-demand therapy. *Haemophilia* 2013; **19**: 808–13.
- Fijnvandraat K, Cnossen MH, Leebeek FWG, Peters M. Diagnosis and management of haemophilia. *BMJ* 2012; **344**: e2707.
- Hazendonk HCAM, Preijers T, Liesner R, Chowdary P, Hart D, Keeling D, Driessens MHE, Gorkom BAPL, van der Meer FJM, Meijer K, Fijnvandraat K, Leebeek FWG, Mathôt RAA, Collins PW, Cnossen MH. Perioperative replacement therapy in haemophilia B: an appeal to “B” more precise. *Haemophilia* 2018; **0**: 1–8.
- Preijers T, Hazendonk HCAM, Fijnvandraat K, Leebeek FWG, Cnossen MH, Mathôt RAA. In silico evaluation of limited blood sampling strategies for individualized recombinant factor IX prophylaxis in hemophilia B patients. *J Thromb Haemost* 2017; **15**: 1737–46.
- Jelliffe RW, Schumitzky A, Bayard D, Milman M, Van Guilder M, Wang X, Jiang F, Barbaut X, Maire P. Model-based, goal-oriented, individualised drug therapy. *Clin Pharmacokinet* 1998; **34**: 57–77.
- Hazendonk HC, Kruip MJ, Mathôt RA, Cnossen MH. Pharmacokinetic-guided dosing of factor VIII concentrate in a patient with haemophilia during renal transplantation. *BMJ Case Rep* 2016; **2016**: bcr2016217069.
- Björkman S. Population pharmacokinetics of recombinant factor IX: implications for dose tailoring. *Haemophilia* 2013; **19**: 753–7.
- Björkman S, Ahlén V. Population pharmacokinetics of plasma-derived factor IX in adult patients with haemophilia B: implications for dosing in prophylaxis. *Eur J Clin Pharmacol* 2012; **68**: 969–77.
- Suzuki A, Tomono Y, Korth-Bradley JM. Population pharmacokinetic modelling of factor IX activity after administration of recombinant factor IX in patients with haemophilia B. *Haemophilia* 2016; **22**: e359–66.
- Neufeld EJ, Solimeno L, Quon D, Walsh C, Seremetis S, Cooper D, Iyer NN, Hoxer CS, Giangrande P. Perioperative management of haemophilia B: a critical appraisal of the evidence and current practices. *Haemophilia* 2017; **23**: 821–31.
- Kiang TKL, Sherwin CMT, Spigarelli MG, Ensom MHH. Fundamentals of population pharmacokinetic modelling: modelling and software. *Clin Pharmacokinet* 2012; **51**: 515–25.
- European Medicines Agency. Guideline on reporting the results of population pharmacokinetic analyses. 2007.
- Lindstrom ML, Bates DM. Nonlinear mixed effects models for repeated measures data. *Biometrics* 1990; **46**: 673–87.
- Anderson BJ, Holford NHG. Mechanism-based concepts of size and maturity in pharmacokinetics. *Annu Rev Pharmacol Toxicol* 2008; **48**: 303–32.
- Bergstrand M, Hooker AC, Wallin JE, Karlsson MO. Prediction-corrected visual predictive checks for diagnosing nonlinear mixed-effects models. *AAPS J* 2011; **13**: 143–51.
- Sherwin CMT, Kiang TKL, Spigarelli MG, Ensom MHH. Fundamentals of population pharmacokinetic modelling: validation methods. *Clin Pharmacokinet* 2012; **51**: 573–90.
- Bonate PL. A brief introduction to Monte Carlo simulation. *Clin Pharmacokinet* 2001; **40**: 15–22.

- 22 Hooker AC, Staatz CE, Karlsson MO. Conditional weighted residuals (CWRES): a model diagnostic for the FOCE method. *Pharm Res* 2007; **24**: 2187–97.
- 23 Thai H-T, Mentré F, Holford NHG, Veyrat-Follet C, Comets E. Evaluation of bootstrap methods for estimating uncertainty of parameters in nonlinear mixed-effects models: a simulation study in population pharmacokinetics. *J Pharmacokinet Pharmacodyn* 2014; **41**: 15–33.
- 24 Brekkan A, Berntorp E, Jensen K, Nielsen EI, Jönsson S. Population pharmacokinetics of plasma-derived factor IX: procedures for dose individualization. *J Thromb Haemost* 2016; **14**: 724–32.
- 25 European Medicines Agency. Withdrawal of the marketing authorisation application for IXinity. 2013.
- 26 Savic RM, Karlsson MO. Importance of shrinkage in empirical bayes estimates for diagnostics: problems and solutions. *AAPS J* 2009; **11**: 558–69.
- 27 Hazendonk HCAM, van Moort I, Fijnvandraat K, Kruip MJHA, Laros-van Gorkom BAP, van der Meer FJM, Meijer K, Peters M, Schutgens REG, Zwaan CM, Driessens MHE, Polinder S, Leebeek FWG, Mathôt RAA, Cnossen MH. The “OPTI-CLOT” trial: a randomised controlled trial on perioperative Pharmacokinetic-guided dosing of CLOTting factor concentrate in haemophilia A. *Thromb Haemost* 2015; **114**: 639–44.
- 28 Björkman S, Shapiro AD, Berntorp E. Pharmacokinetics of recombinant factor IX in relation to age of the patient: implications for dosing in prophylaxis. *Haemophilia* 2001; **7**: 133–9.
- 29 Björkman S. Comparative pharmacokinetics of factor VIII and recombinant factor IX: for which coagulation factors should half-life change with age? *Haemophilia* 2013; **19**: 882–6.
- 30 Ewenstein BM, Joist JH, Shapiro AD, Hofstra TC, Leissinger CA, Seremetis SV, Broder M, Mueller-Velten G, Schwartz BA; Mononine Comparison Study Group. Pharmacokinetic analysis of plasma-derived and recombinant F IX concentrates in previously treated patients with moderate or severe hemophilia B. *Transfusion (Paris)* 2002; **42**: 190–7.
- 31 Alamelu J, Bevan D, Sorensen B, Rangarajan S. Pharmacokinetic and pharmacodynamic properties of plasma-derived vs. recombinant factor IX in patients with hemophilia B: a prospective crossover study. *J Thromb Haemost* 2014; **12**: 2044–8.
- 32 Björkman S, Berntorp E. Pharmacokinetics of coagulation factors: clinical relevance for patients with haemophilia. *Clin Pharmacokinet* 2001; **40**: 815–32.